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INJECTION OF MISCIBLE ETHYLENE DIBROMIDE INTO SOIL TO KILL JAPANESE BEETLE GRUBS AMONG PLANT ROOTS

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Ethylene dibromide dispersed in water for the elimination of grubs of the Japanese beetle (Popillia japonica Newm.) in soil about nursery plants is authorized by the Department of Agriculture to permit their movement outside the regulated area. Either the fumigant is applied in the field before the plants are dug or, after digging, the soil masses around the roots are soaked in water containing the fumigant. This treatment is based on experiments reported by Mason and Chisholm (3). However, its usefulness is restricted by the large volume of water required for the field treatment--about 5,000 to 10,000 gallons per acre--and the water-logged condition of the soil masses. In 1953 an investigation was begun to explore the possibility of injecting the fumigant in more concentrated form, and an injection method was developed which now has been authorized by the Department. This paper describes these studies, and a guide for using this treatment is appended.

Formulations of Ethylene Dibromide

In the original authorized field treatment about 0.4 gram of ethylene dibromide was applied per square foot of soil. A formulation containing this amount of the fumigant in 20 ml. was developed for injection (Chisholm et al. 1). This formulation, known as miscible ethylene dibromide, is given below.

	Percent by weight
Ethylene dibromide	2.5
Tween 20 (polyoxyalkylene derivative of sorbitan monolaurate)	2.5
Isopropyl alcohol (99 percent)	95.0

1/ In this work the authors were assisted by E. H. Siegler (retired), L. B. Parker, W. W. Maines, P. J. McCabe, and W. C. Fest of the Entomology Research Division, and also by V. A. Johnson and others of the Plant Pest Control Division.

It is a clear solution that is miscible in practically all proportions with water. However, owing to circumstances involving patents and other restrictions, it was necessary in 1956 to substitute a mixture containing 83 percent of ethylene dibromide and 17 percent of a light petroleum fraction, known as Dowfume W-85, and modify the formulation:

	Percent by weight
Dowfume W-85	3.1
Tween 20	2.9
Isopropyl alcohol (99 percent)	94.0

Both formulations were equally effective against the grubs.

Injector

A hypodermic syringe equipped with a 3½-inch needle was used in early tests. It had to be filled manually each time it was discharged into the soil, and the open end of the needle often became clogged. There was need for a hand injector with a reservoir or automatic filling device and a nonclogging needle. Fest (2) modified a commercial calking gun for this purpose. This injector was invaluable in the exploratory work, but it is rather large and cumbersome for the injection of small dosages, and there is no visual check on the volume of liquid ejected.

In 1954 a veterinary hypodermic syringe[2/] was found to be more suitable. This syringe is of 2 cc. capacity, graduated in units of 0.1 cc., and has a two-way valve and 48 inches of rubber tubing for automatic filling. Since it is made of glass, the operator can observe the amount of liquid drawn into the chamber and ascertain when all of it is discharged. The liquid is ejected with one down-stroke of the plunger, and the syringe is refilled as the plunger springs back to its resting position. The No. LNR stainless steel needle, 13-gage and 3½ inches long, was modified by sealing the end with solder and boring two or four holes in the side just above this plug with a No. 60 drill. This modification prevents soil from being pushed into the needle when it is inserted into the ground. This injector is light and easily handled, and can be set quickly to discharge the desired amount of liquid.

2/ B-D Cornwall Injector No. 125IV, supplied by Becton, Dickinson and Co., Rutherford, N. J.

Preliminary Experiments

Preliminary experiments were undertaken to find out how much ethylene dibromide was needed to kill third-instar grubs in the soil and to what degree the type of soil, compaction, moisture, temperature, and other factors modified the insecticidal action. Cylindrical masses of soil, 6 to 16 inches in diameter and 8 to 10 inches deep, were built in 1-inch layers within retaining forms of 1/2-inch-mesh hardware cloth lined with burlap, and 25 or more grubs were confined in every two layers. In some experiments the layers were separated by 16-mesh screening; in others the grubs were confined with soil in cages.

Effect of Different Dosages

The miscible ethylene dibromide was injected to a depth of 3 inches into the upper surface of these soil masses at the rates of 0.2, 0.4, 0.8, and 1.6 grams of the fumigant per square foot. Each dosage was distributed to give the equivalent of five injections per square foot. After these masses had been held for 5 days at room temperature, the grubs were transferred to untreated soil for observation. Many of the grubs at that time appeared to be normal. The 0.2-gram treatment was definitely inadequate; many of the grubs within the upper 6 inches appeared to be normal after 38 days. The 0.4-gram dosage killed all grubs at this depth within 21 days, and the 0.8- and 1.6-gram dosages within 14 days. Many grubs in the 6-8-inch layer succumbed to 0.4 gram or more.

It appeared that 0.4 gram of injected ethylene dibromide was about the minimum needed to kill grubs within the upper 6 inches of soil within a reasonable time. As this was the rate used in the field treatments with water-dispersed ethylene dibromide, the elimination of the water did not reduce the effectiveness of the fumigant.

Horizontal Diffusion

To study the horizontal diffusion within the upper 6 inches of soil, 0.08 gram of ethylene dibromide, the amount introduced per injection in the 0.4-gram treatment, was injected 3 inches into wet masses of Penn silt loam at a temperature of 40° F. and after 3, 5, and 7 days the grubs were transferred to untreated soil for observation. When the soil was lightly compacted, an exposure of 3 days was sufficient to kill all grubs within 5 inches of the injection, but when the soil was very compact 7 days were needed. At greater distances the results were variable. To assure that all points within the upper 6 inches have an adequate amount of fumigant, the injections should be not more than 7 inches apart.

Downward Diffusion

To study the downward diffusion, ethylene dibromide was injected to a depth of 3 inches at the rate of 0.4 gram per square foot into wet masses of muck, Sassafras sandy loam, Sassafras loam, Sassafras clay loam, Mentor clay loam, and Penn silt loam at a temperature of 40° F. The dosage was distributed to give the equivalent of five injections per square foot. Seven days later the grubs were transferred to untreated soil for observation.

When the muck was lightly compacted, all grubs to a depth of 8 inches were dead within 14 days. From the rapid mortality at this depth, the treatment was probably effective to 10 or 12 inches. In moderately compacted muck complete mortality was obtained within 14 days only in the upper 4 inches, 21 days were required before all grubs in the 4-6-inch layer succumbed, and 42 days for those in the 6-8-inch layer. In very compact muck only those grubs within the upper 2 inches died within 14 days; it was 35 days before all grubs in the 2-6-inch layer died, and some grubs in the 6-8-inch layer were alive after 49 days.

The type of loam seemed to have little influence on the speed of insecticidal action. Equally good results were obtained with all types. When these soils were lightly compacted, all grubs to a depth of 8 inches were dead within 14 days; when very compact, the grubs in the upper 6 inches died within 21 days, and most of those in the 6-8-inch layer within 42 days.

Temperature

To study the influence of temperature on the insecticidal action within the upper 6 inches, ethylene dibromide was injected into wet compact Sassafras sandy loam at the rate of 0.4 gram per square foot. Seven days later, when the grubs were transferred to untreated soil, 19 percent of those in soil at 35° F. were dead, 44 percent at 45°, 89 percent at 55°, and 98 percent at 65°. Some of the grubs at each temperature survived for several weeks; complete mortality was obtained at all temperatures within 42 days.

In other tests wet muck was subjected to freezing to study its effect on the insecticidal action. The soil became frozen within 12 hours after the injection, and during the following 6 days the temperature went as low as 17° F. All grubs were dead when examined after 7 days. It was evident that freezing was not detrimental to the treatment.

Injection of Plants Before Digging

To determine the effectiveness of the injection treatment when applied in the nursery row, ethylene dibromide was injected into the soil about the base of plants at rates of 0.4 and 0.8 gram per square foot. During the 7-day periods before the plants were dug, the soil temperature usually fluctuated 10 degrees, the average temperatures ranged from 39° to 56° F., and the rainfall from 0.04 to 2.5 inches.

For the treatment of plants to be dug with 12-inch balls, the surface of the soil within 12 inches of the base was cleared of debris and leveled, and then, by means of a template, a pattern of 18 injections was laid out as follows: two $2\frac{1}{2}$ inches from and on opposite sides of the plant, six $4\frac{1}{2}$ inches from the plant and $4\frac{1}{2}$ inches apart, and ten $7\frac{1}{2}$ inches from the plant and $4\frac{1}{2}$ inches apart. For soil balls of different diameter this pattern would be modified. For a 6-inch ball two injections would be made $2\frac{1}{2}$ inches from and on opposite sides of the plant and six injections $4\frac{1}{2}$ inches from the plant and $4\frac{1}{2}$ inches apart. For larger soil balls additional injections would be made in concentric circles 3 inches apart, with the injections in each circle $4\frac{1}{2}$ inches apart. The diameter of the area to be treated should be at least 6 inches larger than that of the soil ball to be dug.

In one series of tests the injections were made to a depth of 3 inches, in another series to 6 inches, and in the third series to 3 and 6 inches alternately. One week after the application the plants were dug and the grubs removed for observation. From 30 to 60 percent of the grubs were found within the upper 5 inches of soil, and some of them 10 inches below the surface. The 0.4-gram dosage killed grubs to a depth of 10 inches. Increasing the dosage to 0.8 gram did not increase the speed of insecticidal action. Equally good results were obtained from injections at both depths. The wetness of the soil seemed to have no effect. The insecticidal action progressed more rapidly at temperatures between 50° and 60° than between 35° and 45° F.

Injection of Balled and Burlapped Nursery Stock

Beginning in 1953 experiments were conducted to establish the effectiveness of this treatment in killing grubs in balled and burlapped nursery stock.

Artificially Infested Stock

The initial experiments were made with balled stock obtained from several commercial nurseries. These soil balls varied greatly in size, shape, type and compaction of the soil, and in the method of wrapping. As the infestation of these plants was negligible, ten or more third-instar grubs were introduced into each ball at depths up to 6 inches a

few days before the treatment was applied. Ethylene dibromide was injected 3 inches into the upper surface of these soil masses at the rate of 0.4 gram per square foot. One injection was made in the 6-inch, two in the 8-inch, three in the 10-inch, four in the 12-inch, five in the 14-inch, and seven in the 16-inch balls. The treatment killed most of the grubs in the upper 6 inches within 2 weeks, but some of them did not succumb for 6 weeks or more.

Naturally Infested Stock

Although the treatment of this artificially infested stock was successful, further information was needed on performance against natural infestations throughout the normal spring and fall shipping seasons at various localities in the infested area. In the spring of 1954 experimental nursery blocks of California privet (Ligustrum ovalifolium) were established in North Carolina, Virginia, Maryland, New Jersey, and Ohio. When it was found early in September that the natural infestation in these blocks was light, 25 third-instar grubs were introduced into the soil about the base of each plant. These grubs had ample opportunity to adjust themselves to their environment before any plants were dug. Beginning in October, at intervals during the fall and spring plants were dug with compact soil balls 12 inches in diameter and 10 to 12 inches deep and wrapped with burlap in the usual way.

The 0.4-gram treatment was applied to the soil balls immediately after wrapping with burlap. At Moorestown, N. J., the balls were held in a chamber in which the soil temperature at the time of digging was maintained. At the other localities they were protected from wind and rain but were subjected to the normal fluctuations of the outdoor temperatures. Seven days later the grubs were transferred to untreated soil for observation.

The effectiveness of this treatment at the various localities is summarized in table 1. The injection of ethylene dibromide practically eliminated the grubs at all of these localities in spite of the differences in the type and condition of the soils. Of the 2,467 grubs recovered, only 14 had not succumbed at the end of 6 weeks. Most of the grubs were within the upper 8 inches, but an occasional grub was found 10 to 12 inches below the surface.

It was suspected that the few grubs that did not succumb within 6 weeks had been near the bottom of the soil balls, where the concentration of the fumigant might not have been adequate. In another series of experiments ethylene dibromide was injected into the balls to depths of 3 and 9 inches. This introduced the fumigant at the rate of 0.8 gram per cubic foot. The results of these tests are also summarized in table 1. Of the 5,062 grubs recovered, only 22 had not succumbed at the end of 6 weeks. Since the fumigant was distributed throughout the soil masses, it would appear that these few grubs were more resistant to the insecticide.

There are advantages in applying the treatment on a volumetric basis, rather than according to the area of the upper surface, because one can adjust the dosage to the depth and thus avoid overdosing the shallow balls and assure an adequate concentration in the deeper ones. It does not seem practical at this time to apply the treatment to balls greater than 12 inches in depth because of the large number of injections required. To assure the distribution of the fumigant throughout the soil mass, injections should be made to a depth of 3 inches when the ball is less than 8 inches deep and to 3 and 9 inches when it is more than 8 inches deep.

Effect of Soil Conditions

The influence of temperature, water content, and organic matter on the insecticidal action of ethylene dibromide in the soils of the experimental nurseries is summarized in table 2. The insecticidal action was slower at low temperatures, and in soils high in organic matter, but the water content of the soil did not seem to be a limiting factor. Within 6 weeks practically all the grubs were destroyed in these soils.

Injection of Potted Plants

In 1954 and 1955 experiments were conducted to determine the effectiveness of injected ethylene dibromide in killing third-instar grubs in the soil of potted plants. Pots ranging in diameter from 2 to $10\frac{1}{2}$ inches were used. The rate of injection ranged from 0.1 to 1 gram per cubic foot. To facilitate the applications, the miscible formulation was diluted with water to give the desired dosage in 1 to 16 ml., according to the size of the pot. The number of injections per pot ranged from one in the 2- and 3-inch pots to four in the 8- and $10\frac{1}{2}$-inch pots. Injections were made to half the depth of the soil, except in the two largest pots, where they were made to a depth of 3 inches. After treatment the pots were left undisturbed for 7 to 10 days, and then the grubs were transferred to untreated soil. The results, summarized in table 3, show that the 0.4-gram dosage eliminated the grubs within 4 weeks.

The influence of the volume and type of soil and the temperature on the insecticidal action of ethylene dibromide in the soil of potted plants is summarized in tables 4, 5, and 6. The rate of insecticidal action was about the same in the 2-inch and the $10\frac{1}{2}$-inch pots, indicating that the volume of the soil did not limit the effectiveness of the fumigant. Neither did the type of soil or temperature seem to be important limiting factors.

Effect on Plants

To obtain information on the tolerance of plants to injections of ethylene dibromide into the soil about their roots, tests were undertaken in cooperation with 45 commercial nurseries and 9 greenhouse establishments within the area infested by the Japanese beetle. The growers selected the plants to be tested and kept them on their premises until the reactions to the treatment had been determined. Tests were made with both balled nursery stock and potted plants.

Balled Nursery Stock

The plants were dug, wrapped in burlap, and prepared for shipment according to the usual practice at the establishment. Ethylene dibromide was injected into the soil at the rate of 0.8 gram per cubic foot. Most of the tests were made during the normal fall and spring shipping seasons, but some of them were made late in the spring when the plants were growing vigorously and were more susceptible to mechanical and chemical injury. After the treated plants had been held for about a week in a shed or on a loading platform, they were set out on the nursery grounds where they received no more than normal care. They were observed periodically during the following 3 to 5 months. The reaction was considered to be satisfactory when the treated plants could not be distinguished from the untreated plants and unsatisfactory when they were retarded in growth, of abnormal color, or killed.

Abelia, Azalea, Buxus, Chamaecyparis, Chrysanthemum, Cornus, Gaillardia, Iberis, Ilex, Juniperus, Ligustrum, Lonicera, Malus, Nandina, Phlox, Picea, Pieris, Pinus, Polemonium, Rhododendron, Spiraea, Thuja, and Viburnum were usually tolerant to the treatment. The reaction of Taxus was satisfactory in some tests but not in others. Acer, Daphne, Pseudotsuga, and Syringa were seriously injured or killed. The reactions of Castanea, Kalmia, and Magnolia were in doubt because of the poor condition of the untreated plants.

Potted Plants

Ethylene dibromide injected into the soil of potted plants at the rate of 0.4 gram per cubic foot caused no serious injury to Ageratum, Azalea, Begonia, Gardenia, Hydrangea, Ilex, and Rosa, but retarded or killed Daphne, Pelargonium, and Viola.

An attempt was made to lessen the phytotoxicity by diluting the miscible formulation with water and injecting a large volume or by introducing a soluble fertilizer at the same time as the fumigant. However, on a susceptible plant such as Pelargonium there was no indication that the injury had been reduced. Several other alcohols were tested

as substitutes for isopropyl alcohol in the formulation. The reactions of plants to formulations containing methyl or ethyl alcohol were about the same as to that containing isopropyl alcohol. Formulations containing n-butyl, isobutyl, or amyl alcohol were definitely more toxic.

Summary

The ethylene dibromide treatments authorized to permit movement of nursery plants out of areas regulated on account of the Japanese beetle (Popillia japonica Newm.) are restricted in usefulness because a large volume of water is required. A method of injecting the fumigant in more concentrated form has been developed and authorized.

A miscible formulation containing 2.5 percent of ethylene dibromide was injected into the soil with a veterinary hypodermic syringe with a stainless steel needle, modified to prevent soil from being pushed into it.

Injection at the rate of 0.4 gram per square foot killed grubs in soil about the roots of plants in the nursery row, 0.4 gram per cubic foot in the soil of plants growing in pots and other containers, and 0.8 gram per cubic foot in balled and burlapped nursery stock.

The insecticidal action was slower at low temperatures, in soils high in organic matter, and in very compact soils, but the water content of the soil did not seem to be a limiting factor. Under some conditions 6 weeks or more elapsed before all the grubs succumbed to the treatment.

Many kinds of plants in the commercial nurseries and greenhouses were usually tolerant to the treatment, but some of them were seriously injured or killed.

Literature Cited

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1954. Hand injector of liquid soil insecticides. U. S. Dept. Agr. Ent. Res. Branch ET-314, 4 pp.

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Table 1.--Effectiveness of injected ethylene dibromide against third-instar Japanese beetle grubs in balled and burlapped stock at different localities

Locality	Number of grubs recovered	Percent mortality after--					
		1 week	2 weeks	3 weeks	4 weeks	5 weeks	6 weeks
0.4 gram per square foot to 3 inches							
Faison, N. C.	633	84	95	97	98	99	99
Moorestown, N. J.	959	75	95	97	98	99	99+
Mt. Vernon, Md.	391	57	92	98	99	100	100
Norfolk, Va.	206	43	59	90	95	95	100
Penrose, N. C.	80	59	93	93	98	99	99
Perry, Ohio	198	86	100	100	100	100	100
Total or average	2,467	72	92	97	98	99	99+
0.8 gram per cubic foot to 3 and 9 inches							
Moorestown, N. J.	2,439	37	95	98	99	99	99+
Mt. Vernon, Md.	955	61	84	90	95	97	99
Norfolk, Va.	1,230	47	76	86	96	98	99+
Penrose, N. C.	158	66	100	100	100	100	100
Perry, Ohio	280	52	87	96	97	100	100
Total or average	5,062	52	87	94	97	99	99+

Table 2.--Influence of temperature, moisture, and organic matter on the effectiveness against Japanese beetle grubs of ethylene dibromide injected into balled nursery stock

Factor	Number of grubs recovered	Percent mortality after--					
		1 week	2 weeks	3 weeks	4 weeks	5 weeks	6 weeks
Temperature of soil (°F.):							
33-39	1,659	22	85	95	97	98	99
40-49	2,252	45	85	91	96	98	99
50-59	2,166	72	88	96	99	99	99+
60-69	1,303	89	98	99	99	99+	99+
70-79	129	81	92	100	100	100	100
Saturation by water (percent):							
0-20	692	67	86	96	98	98	99+
40-60	4,945	61	88	94	97	98	99+
80-100	1,892	40	90	97	99	99	99+
Organic matter:							
Low	5,146	59	92	97	98	99	99
Moderate	716	67	94	98	99	99+	99+
High	1,667	44	73	88	96	99	99+

Table 3.--Effectiveness of various dosages of ethylene dibromide injected into potted plants against third-instar Japanese beetle grubs

Grams per cubic foot	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	3 weeks	4 weeks
0.1	300	17	79	94	97
.2	2,393	43	89	97	99
.4	3,910	60	98	99+	100
.6	1,470	--	99	100	100
.8	2,529	47	99	100	100
1.0	1,470	--	99+	100	100

Table 4.--Influence of the volume of soil in pots on the effectiveness of various dosages of injected ethylene dibromide against third-instar Japanese beetle grubs

Diameter of pots (inches)	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	3 weeks	4 weeks
		0.2 gram per cubic foot			
2	404	45	88	95	97
3	705	37	82	96	99+
4	517	50	84	94	97
8	767	42	94	99	99
		0.4 gram per cubic foot			
2	891	52	95	97	100
3	1,197	45	98	99+	100
4	1,019	80	99	99+	100
8	353	67	99	100	100
$10\frac{1}{2}$	450	--	100	100	100
		0.8 gram per cubic foot			
2	480	98	100	100	100
3	800	39	97	100	100
4	495	--	100	100	100
8	304	55	100	100	100
$10\frac{1}{2}$	450	--	100	100	100

Table 5.--Influence of type of soil in pots on the effectiveness of various dosages of injected ethylene dibromide against third-instar Japanese beetle grubs

Type of soil	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	3 weeks	4 weeks
	0.2 gram per cubic foot				
Sassafras sandy loam	1,308	45	91	98	99
Penn silt loam	512	59	98	99	100
Muck	573	43	89	97	99
	0.4 gram per cubic foot				
Sassafras sandy loam	1,497	65	99	99	100
Penn silt loam	1,184	69	99	99+	100
Muck	1,229	44	97	99+	100
	0.6 gram per cubic foot				
Sassafras sandy loam	490	--	100	100	100
Penn silt loam	490	--	98	100	100
Muck	490	--	99	100	100
	0.8 gram per cubic foot				
Sassafras sandy loam	1,041	45	99+	100	100
Penn silt loam	746	52	99+	100	100
Muck	742	52	96	100	100
	1.0 gram per cubic foot				
Sassafras sandy loam	490	--	100	100	100
Penn silt loam	490	--	9	100	100
Muck	490	--	100	100	100

Table 6.--Influence of the temperature of the soil in pots on the effectiveness of various dosages of injected ethylene dibromide against third-instar Japanese beetle grubs

Mean temperature (°F.)	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	3 weeks	4 weeks
		0.2 gram per cubic foot			
33	161	14	89	93	100
35	614	30	93	99	99
36	143	19	97	99	100
40	103	97	100	100	100
50	1,322	49	85	95	98
		0.4 gram per cubic foot			
33	160	9	96	99	100
35	204	43	99	100	100
36	148	21	99	100	100
40	101	100	100	100	100
50	48	96	100	100	100
66	1,329	68	97	99+	100
68	1,920	--	98	100	100
		0.8 gram per cubic foot			
33	155	92	100	100	100
35	206	34	100	100	100
36	150	9	100	100	100
40	50	96	100	100	100
50	48	100	100	100	100
68	1,920	--	98	100	100

Treatment Guide

The dosage of ethylene dibromide injected into the soil must be carefully controlled to assure that sufficient fumigant is introduced to kill the grubs but not enough to injure the roots of the plants. A guide is therefore given for treatment in the nursery row, and injection into soil of balled and burlapped stock and potted plants.

Injection in the nursery row.--Treat an area at least 6 inches larger in diameter than the ball to be dug. Inject 2 ml. of miscible ethylene dibromide to a depth of 3 inches at each of the following points: two injections $2\frac{1}{2}$ inches from and on opposite sides of the plant, six $4\frac{1}{2}$ inches from the plant and $4\frac{1}{2}$ inches apart, and additional injections as needed in concentric circles 3 inches apart, spacing the injections $4\frac{1}{2}$ inches apart in each circle.

Injection of balled and burlapped stock.--Determine the horizontal diameter and the depth of the soil ball. For balls less than 10 inches in diameter use 2 ml. and for larger balls 4 ml. per injection. Inject the fumigant to a depth of 3 inches, using the number and arrangement of injections given in table A.

Injection of plants in pots or other containers.--Determine the volume of soil in the container, and dilute the ethylene dibromide formulation with water and make the number of the injections indicated in table B. Make the injections to a depth of 3 inches, except when the soil is less than 6 inches deep, and then to one-half the depth of the soil.

CAUTION.--The injection of ethylene dibromide into soil close to the roots of a living plant in sufficient amount to kill a grub may cause some injury to the plant. A grower should test the susceptibility of his plants before undertaking the injection of ethylene dibromide on a large scale.

Table A.--Injection into balled nursery stock

Size of ball (inches)		Number of injections	
Horizontal diameter	Depth	Top	Bottom
	2 ml. per injection		
4-6	4-6	2	0
6-8	4-6	4	0
	6-8	5	0
8-10	4-6	5	0
	6-8	7	0
	8-10	5	4
	4 ml. per injection		
10-12	4-6	4	0
	6-8	5	0
	8-10	4	3
	10-12	4	4
12-14	4-6	5	0
	6-8	7	0
	8-10	5	4
	10-12	6	6
14-16	4-6	7	0
	6-8	9	0
	8-10	6	6
	10-12	7	7
16-18	4-6	9	0
	6-8	12	0
	8-10	8	7
	10-12	9	9
18-20	4-6	11	0
	6-8	15	0
	8-10	9	9
	10-12	11	11
20-22	4-6	13	0
	6-8	18	0
	8-10	11	11
	10-12	13	13
22-24	6-8	21	0
	8-10	13	13
	10-12	16	16

Table B.--Injection into soil of plants growing in pots or other containers

Volume of soil	Milliliters of formulation made up to 1 pint with water	Number of injections
	1 ml. per injection	
Fluid ounces		
2	19	1
4	38	
8	80	
12	118	
16	156	
20	199	
Quarts		
3/4	118	2
1	159	
1 1/4	199	
1 1/2	118	4
1 3/4	138	
2	159	
2 1/4	177	
2 1/2	199	
2 3/4	218	
	2 ml. per injection	
Quarts		
3	118	4
3 1/2	138	
4	158	
5	198	
6	237	
7	277	
8	316	
9	237	6
10	263	
12	316	
14	369	
16	316	8
18	356	
20	395	